

MYCOLOGICAL, BIOCHEMICAL AND HISTOPATHOLOGICAL STUDIES ON ACUTE FUSARIOTOXICOSIS IN SHEEP

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ABSTRACT

The present study was conducted on one hundred affected sheep which were collected from desert districts in governorates of (Giza; 6th. October and El-Wadi-El-Gadid). Sixty percent of the collected sheep sera had a mean levels of T-2, zearalenone and fumonisins (2.5 ± 0.2 , 4.3 ± 0.5 and 25.0 ± 2.0) respectively. The used feeds and underground water in breeding of this sheep were examined mycologically which revealed that all examined samples gave a variable severity of pollution. Seven genera and 15 species of fungi were recovered from feeds and water. The most predominant isolates belong to members of genus *Aspergillus* with an average of (5-100%), followed by *Fusarium* spp. (40-90%), *Penicillium* spp. (10-55%) and *Mucor* spp. (10-50). The *Fusarium* toxins were detected in the same feed samples, the highest amount was estimated in crushed yellow corn (60%) namely FB1, T2 and zearalenone with the mean levels of (48.4 ± 1.0 ; 3.0 ± 0.1 and 0.84 ± 0.03) respectively. The significant high levels of FB1 in the present feed samples and serum of diseased sheep gave a large possibility that FB1 was responsible for this disease outbreak in sheep. On the other hand, the biochemical examination of diseased sheep sera for estimation of toxic effects was based on the assumption that the elevated activities in levels of serum enzymes such as AST, ALT, GGT, LDH and urea (Table, 5). Slightly decrease in ceratinine, calcium and phosphorus levels comparing with the apparently healthy group. The pattern of protein electrophoresis showed a significantly decrease values in were recorded serum total protein, alpha globulin, beta globulin with slightly increase in gamma globulin. The internal organs of dead cases had various significant pathological changes in vital organs including hemorrhagic, alveolar pneumonia and calcification in lung. The liver showed hemorrhage, oedema, vacuolar degeneration and necrosis of hepatocytes with evidence of preneoplastic stage in liver cells. Whereas, the kidney showed vacuolar degenerating changes and necrosis of the tubular epithelium, in addition to glomerular oedema and calcium deposition. Significant dangerous effect of environmental pollutions particularly fusarium species and their toxins were recorded in the study.

Keywords: pollution; biochemical alterations; fusarium

INTRODUCTION

The increased importance of animal production due to progressive elevated requirement of human consumption gave an intensive attention of animal health status. The environmental pollution is considered the essential cause of animal diseases particularly pollution with fungi and their toxins for the used feed and water in animal breeding and elsewhere, contamination of human food. Mycotoxins are a group of structurally diverse, mold elaborated compounds that induce diseases known as mycotoxicosis in humans and animals. As much as twenty-five percent of the world's food crops are estimated to be contaminated with mycotoxins. Ingestion of sufficient quantities of mycotoxin-contaminated material leads to acute, and more commonly, chronic intoxication (**Hassan et al., 2003 and 2009**).

The mycotoxins of greatest agricultural and public health significance include aflatoxins, ochratoxins, trichothecenes, fumonisins, zearalenone, and ergot alkaloids (**Hassan et al., 2004, 2008 and 2009**). However, the fungi of *Fusarium* species and their toxins are widely distributed through the world where they occur in soil, on plants, plants debris and similar organic substrates. They cause significant economic losses in agriculture, morbidity and mortality in animals and immunological compromised humans, where it is capable of killing cells by causing extensive damage to cellular membrane (**Ajello and Hay, 1998 ; Mogda, Mansour, et al., 2002**). On the other hand, epidemiological studies associated with fusarium toxins had a wide range of biological effects, including pulmonary oedema in pigs and ruminants (**Harrison et al., 1990**), nephrotoxicity and liver cancer in rats (**Gelderblom et al., 1996**). Although, its effects on human are difficult to be determined.

Fumonisin B₉ had been statistically associated with a high incidence of oesophageal cancer in certain areas of Transkei, South Africa and also in China (**Chu and Li, 1994**). The International Agency for Research on Cancer has declared *F. moniliforme* form toxins as potentially carcinogenic to human. **Gelderblom et al. (1994)** proposed that FB₁ was a tumour promoter at doses not causing significant liver pathology but when given at overtly hepatotoxic dose, it was also a weak initiator. Also, the lymphocytes decreased in response to Zearalenone especially

for LD50 dose. Many data showed that this mycotoxin induced immunosuppression in depressing T or B lymphocyte activity (**Berek et al., 2001**). All the previous literatures recorded that the pollution affect upon the growth rate and health of human being and animals including aneamia , stunted growth , carcinogenic, tremorgenic, haemorrhagic, dermatitic, pulmonary edema, immunosuppressive and hormonal effects (**Hassan, 1998 and 2003 ;and Hassan et al., 2003 ;2004;2008 and 2009**). Whenever, sheep breeding and their production is the main source of food for human in the desert districts. So, the aim of the present work was to investigate the problem of fungal and fusarium mycotoxins pollution of feed and underground water and its role in the health status of sheep at some deserts Governorates (Giza, El-Wadi El Gadid and 6 th October).

MATERIAL AND METHODS

Material

Samples

Serum, feed and water samples

Blood samples were collected from one hundreds diseased cases of sheep at desert districts in governorates of Giza; 6th October and El-Wadi-El-Gadid were investigated. The cases of sheep suffered from loss of weight gain, low productivity, diarrhea, mastitis, disturbance in fertility and sudden mortality of some cases. From districts of diseased cases, 100 samples of sera, 150 feeds and 20 samples of underground water which used in breeding of diseased sheep were collected. The samples of feed and water were collected in sterile plastic container to prevent any contamination.

Internal organs

From the recently deed cases of animal from disease outbreak, the internal organs were collected and imbedded in 10% formalin solution for further histopathological examination. These organs included liver, kidney, lung, bronchial lymph node and heart.

Mycotoxins standards

Standers and immunoaffinity column of Zearalenon, T2 and FB1, were purchased from Sigma Chemical Company (USA).

Methods

Mycological examination of samples

The samples of feeds and underground water which were consumed by symptomatically affected sheep were subjected for isolation and identification of fungi as recommended by **(Conner et al., 1992)**.

Detection of mycotoxins in feed and sera of diseased sheep

Detection of mycotoxins in serum of sheep and feed stuffs by fluerometric methods as described by **Hansen (1993)** using immune-affinity column method.

Biochemical investigations of sheep sera:

Blood samples were collected in small labeled dry and clean vials without anticoagulant in centrifuge tube, allowed to clot and then centrifuged at 3000 rpm for 15 minutes for separation of serum which used to assay the biochemical parameters The biochemical assays of serum gamma glutamyle transferase (GGT) and lactic dehydrogenase (LDH) activities were determined according to methods of **(Szase et al., 1976)** ,aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities according to **Reitman and Frankel (1957)**, serum urea according to **Wybenga et al. (1971)**, serum creatinine level according to **Henry (1974)**, Estimation of serum total protein and electrophoretic pattern were carried out after **SonnenWirth and Jaret (1980)** ; **Davis (1964)** , respectively.

However, measurement of calcium, ph. and Mg. were carried out as the technique described in the references **(Brown et al., 1986 ; Brown and Taylor, 1995)**.

Histopathological studies

Tissue specimens were collected directly from lung, bronchial lymph node, heart, liver, spleen, kidneys and intestine of freshly dead cases for histopathological examination. They were kept in 10% neutral buffered formalin for at least 24 hours, routinely processed by the standard paraffin embedding technique and stained with Hematoxylin and Eosin for routine histopathological investigation. Prussian blue stain for detectimy hemosidrin pigments **(Bancroft et al., 1994)**.

Statistical Analysis

The obtained data were computerized and analyzed for significance, Calculation of standard error and variance according to (SPSS 14, 2006).

RESULTS AND DISCUSSION

The economical importance of sheep animals in desert districts Governorates were at the top to other part in Egypt, where, peoples in these districts their life depend on its products such as meat, milk, wool and leather obtained from these animals (Agaoglu, 1991; Camas et al., 1994 ; Hassan et al., 2008)

In the present study, the current data in table (1) showed that, sera of one hundred cases of diseased sheep outbreaks which suffered from loss of weight gain, low productivity, diarrhea, mastitis, disturbance in fertility and sudden mortality of some cases at desert districts in governorates of Giza; 6thOctober and El-Wadi-El-Gadid, contained significant levels of fusarium toxins. Meanwhile, sixty percent of these sheep had the mean levels of fusarium toxins as T-2, zearalenone and fumonisins (2.5±0.2, 4.3±0.5 and 25.0±2.0) respectively. The results indicated that serum of diseased sheep contained higher mean significant level of FB1 than other types of fusarium toxins which suggested being the essential cause of disease. Mycotoxins in sera of sheep and cattle in Egypt in association with symptoms of toxicities were previously reported by Hassan (1994); Hassan et al. (2003; 2004 and 2009).

Table (1): Determination of fusarium toxins in serum of diseased sheep

Animals	Prevalence of fusarium toxins			Mean levels of fusarium toxins (ppm)		
	No. of tested	No. of +ve	%	Fumonisin	T-2	Zearalene
Sheep	100	60	60	25.0±2.0	2.5±0.2	4.3±0.5

- Results are expressed as means ± SEM, student 't' test

The effects of fusarium toxins in human and animals were varied from carcinogenic and nephrotoxic and immunosuppressive health effects (Morriss, et al, 1997). Although the main route of human exposure to mycotoxins has been identified as the direct ingestion of contaminated cereals and grains (Morriss, et al, 1997), there are many studies about whether the ingestion of meat, milk, and eggs originating from mycotoxin-exposed food-production animals is a significant pathway for mycotoxins among humans (Hassan et al., 1997; Wafia, H. Abdallah and Hassan, 2000 ; Hassan et al., 2004 and 2009). The search focused to recovered the accurate causes and sources of this collected cases, therefore, the direct factors to the animal consumption were examined .The fungal examination for feeds , feedstuffs and underground water (which the only available source of water in these districts), revealed that all examined samples gave a variable rates of pollution. Seven genera and 15 species of fungi were isolated from consumed foods and drinking water. The most predominant isolates belong to members of genus *Aspergillus* with a range of (5-100%), followed by *Fusarium* spp. with (40-90%), *Penicillium* spp. (10-55%) and *Mucor* spp. (10-50%).

Whereas, the frequency of isolation of other spp. as *Rhizopus* spp., *C.albicans* and *Rhodotorula* spp. were relatively low. On the other hand, the fungal contamination of underground water was significantly high as compared with standard healthy water which must be free from any signs of pollution (Table, 2). However, *F.moniliform*, *F.oxysporum* and *F. solani* were the most frequent isolated members of *Fusarium* from feed samples (Table, 3). The fungus of *F.moniliform* was recovered from all examined feed samples with(20-65%), while, *F.oxysporum* was isolated from lower examined samples (5-10%) with exception of wheat straw samples .Whereas, the species of *F. nival* and *F. fusaroides* were only isolated from (Soya bean meal and crushed yellow corn), respectively with the same rate (5%). It is clear from the result that crushed yellow corn and wheat straw were the most contaminated materials followed by hay, Soya bean and drawa. While, the underground water was the lowest contaminated samples. These differences in the level of contamination may be due to the exposure of the examined samples to different climatic condition either during preparation or transportation or storage. These

findings were in agreement with the results of (Hassan et al. 2003; 2004; 2008 and 2009), who recovered most of these fungi from the examined feed and water samples.

Table (2): Prevalence of fungi in feeds and underground water used for breeding of sheep

Fungal Species	Crushed yellow corn(30)		hay(35)		Wheat straw(20)		Soya bean meal(35)		Drawa (Leaves of yellow corn) (30)		Underground water (20)	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
<i>Aspergillus sp.</i>	20	100	19	95	20	100	15	75	10	50	1	5
<i>A. flavus</i>	18	90	17	85	18	90	7	35	40	20	1	5
<i>A. niger</i>	16	80	15	75	15	75	14	70	36	18	10	50
<i>A. candidus</i>	1	5	--	--	--	--	2	10	30	15	0	0
<i>A. fumigatus</i>	4	20	7	35	--	--	2	10	20	10	1	5
<i>A. ochraceus</i>	5	25	19	5	1	5	1	5	16	8	0	0
<i>A. terrus</i>	5	25	2	10	3	15	3	15	10	5	0	0
<i>Fusarium sp.</i>	10	50	18	90	15	75	8	40	8	40	0	0
<i>Penicillim sp.</i>	7	35	9	45	6	30	10	50	11	55	2	10
<i>Mucor sp.</i>	10	50	6	30	2	10	10	50	3	15	0	0
<i>Rhizopus sp.</i>	1	5	1	5	3	15	4	20	1	5	0	0
<i>C.albicans</i>	2	10	0	0	0	0	1	5	2	10	1	5
<i>Rhodotorula sp</i>	1	5	0	0	1	5	0	0	2	10	2	10

Table(3):Prevalence of fusarium species in feeds of sheep suffering from problems of animal diseases from different districts at el Wadi El Gedid

Fusarium Species	Crushed yellow corn		Hay		Wheat straw		Soya bean meal		Drawa (Leaves of yellow corn)	
	No.	%	No.	%	No.	%	No.	%	No.	%
<i>F.moniliforme</i>	4	20	13	65	8	40	6	30	7	35
<i>F.oxysporum</i>	1	5	1	5	-	-	1	5	2	10
<i>F.solani</i>	1	5	1	5	4	20	-	-	-	-
<i>F.sporotrichoides</i>	1	5	-	-	1	5	-	-	-	-
<i>F. aquaeductum</i>	1	5	-	-	1	5	-	-	-	-
<i>F. nival</i>	-	-	-	-	-	-	1	5	-	-
<i>F. fusaroides</i>	1	5	-	-	-	-	-	-	-	-
<i>F. equiseti</i>	-	-	-	-	1	5	-	-	-	-
<i>F. tricinctum</i>	1	5	3	15	-	-	-	-	-	-

When, the feed samples which contaminated with fusarium spp. were subjected for detection of Fusarium toxins, the results revealed that the largest amount was detected in crushed yellow corn (60%) namely FB1, T2 and zearalenone with the mean levels of (48.4±1.0; 3.0±0.1 and 0.84±0.03 ppm), respectively.

It was interesting to report here that the samples of wheat straw contained only FB1 at a rate of (70%) with a mean level of (20±0.9 ppm) (Table, 4). The significant levels of FB1 in the present feed samples and serum of diseased sheep gave a large possibility that FB1 was responsible for the disease outbreak in sheep. The Food and drug administration has established recommended maximum levels for aflatoxins and fumonisins in animal feed. For swine, ruminants including sheep, and poultry, the recommended maximum levels of total fumonisins in complete feeds are 10, 30, and 50 µg/g, respectively (FDA, 1994). Therefore, the detected levels of FB1 were significantly

over the permissible limits in feeds particularly FB1 toxin in examined sheep feed samples which ranged from (15.0±0.2-48.4±1.0 ppm). The same findings were detected by many authors as (**Hassan et al., 2002; 2003;2004; 2008 and 2009**) ; **El-Hamaky (2001)** ; **El Ahle et al.(2006)**

Table (4): Detection of fusarium toxins in feeds.

Fusarium Species	Prevalence of fusarium toxins			Mean levels of fusarium toxins (ppm)		
	No. of tested	No. of +ve	%	Fumonisin	T-2	Zearalenone
Crushed yellow corn	10	6	60	48.4±1.0	3.0±0.1	0.84±0.03
Hay	10	5	50	17.0±1.3	-	0.71±0.0
Wheat straw	10	7	70	20±0.9	-	-
Soya bean meal	10	4	40	15.0±0.2	2.0	0.99±0.005
Drawa (Leaves of yellow corn)	10	4	40	27.0±3.22	1.0±0.01	1.50±0.0

- Results are expressed as means ± SEM, student 't' test

On the other hand, the biochemical examination of affected sheep sera for estimation of toxic effects was based on the elevated activities in levels of serum enzymes activities such as (AST, ALT, GGT, LDH) and urea concentration in Table, (5). While, a slightly decreases in ceratinine level compared with the apparently healthy group. These results reflected the damage in the different organs (**Cheng et al., 2001 ; Asrani, et al., 2006**). Increased serum enzymes activity was observed by feeding toxic diets due to hepatic degeneration and subsequent leakage of enzymes into circulation. (**Chen et al., 2008 ; Wang et al., 2008**). It is reported that the significant effect of fusarium toxins are the alteration in serum concentration of kidney and liver enzymes ,total protein, albumin, minerals and lipid profiles (**Kubena et al., 1997 ; Mogdaet al., 2002**).

Table (5); Biochemical parameters in serum of diseases sheep cases at desert districts in comparison to healthy cases.

Parameter	Apparently healthy	Diseased group
AST u/l	53.67±4.91	124.9***±7.94
ALT u/l	40.66±2.18	93.6***±5.48
GGT u/l	97.57±1.38	111.56*±5.11
LDH u/l	718.4±22.36	811.0*±24.11
urea mg%	41.11±2.15	53.52**±3.81
Creatinin mg%	1.31±0.07	0.9±0.14*
Uric acid mg%	3.17±0.37	5.1**±0.34
Calcium mg%	9.22±0.33	7.46**±0.41
Phosphorus mg%	6.31±0.32	5.77±0.17

Results are expressed as means ± SEM ,student ‘t’ test

The high concentrations of serum urea in sheep fed on contaminated diet may be leading to increased ammonia absorption caused by altered protein turnover in the rumen micro-flora, or altered protein metabolism in sheep tissues. In ruminants, serum urea levels are affected by protein digestion and metabolism by the rumen biomass. A large portion of dietary protein is hydrolyzed and deaminated by rumen micro-flora, giving rise to peptides and free ammonia in the rumen (**Herdt, 2000**). A part of free ammonia is absorbed and is metabolized to urea in the liver. If microbial protein synthesis in the rumen which is inhibited by mycotoxins, more free ammonia remains in the rumen, is absorbed into the blood, and is metabolized to urea, resulting in elevated blood urea concentrations. **Danicke et al. (2005)** observed that postprandial rumen fluid ammonia concentrations were consistently higher when *Fusarium* mycotoxin-contaminated wheat was fed to sheep. Inhibition of protein synthesis results in elevated concentrations of free Amino acid that are used for energy utilization, resulting in increased serum urea. The results of the present study are in agreement with those of **Chowdhury and Smith (2004)**, who observed that excessive serum concentrations of uric

acid in laying hens were a result of feeding feedborne *Fusarium* mycotoxins. Moreover, in a subsequent study with laying hens, they found that feeding contaminated grains led to reduced hepatic fractional protein synthesis rates (**Chowdhury and Smith, 2005**). **Danicke et al. (2006)** also observed a reduction in fractional protein synthesis rates in the kidneys, spleen, and ileum of pigs exposed to DON.

At the same time concentrations of serum calcium and serum phosphorus were decreased due to feeding *Fusarium* mycotoxin-contaminated diets. This results were agree with **Díaz and Smith (2006)**. *Fusarium* inducing significantly decreased values in serum total protein, alpha globulin, beta globulin and while slightly increase in gamma globulin, these results agree with (**Rotter et al., 1994**).

Table (6); Patterns of protein electrophoresis in serum of diseases sheep cases at desert districts in comparison to healthy cases (mg/dl).

Parameter	Apparently healthy	Diseased group
Alb	2.35±0.12	1.87**±0.07
T.alpha	0.96±0.1	0.87±0.09
Alpha1	0.41±0.03	0.4±0.02
Alpha1	0.55±0.02	0.47*±0.02
t. beta globulin	1.09±0.04	1.02±0.03
Beta1	0.5±0.02	0.55±0.04
Beta2	0.59±0.01	0.47*±0.04
Gamma1	1.59±0.11	1.53±0.05
Gamma2	0.34±0.03	0.52±0.03
Gamma globulin	1.93±0.15	2.05±0.1
T.globulin	3.98±0.33	3.94±0.29
A/G ratio	0.59±0.03	0.43**±0.03
T. protein	6.33±0.55	5.81±0.08

The globulin component (Table, 6) showed drop in α_1 , α_2 and β_2 globulin in all the experiment while decrease γ_1 globulin. This may be attributed to that *Fusarium* fungi cause's hepatotoxic, nephrosis, hemorrhages in (liver and kidneys) (Tietz, 1996) *Fusarium* mycotoxins might affect the synthesis of globulins of hepatic origin as well as globulins of lymphoid origin. Rotter et al. (1994) suggested that *Fusarium* mycotoxins can directly affect α -globulin synthesis in the liver. In addition, *Fusarium* fungi has immunosuppressive effect inhibit nearly cellular and humeral immunologic reaction have been reported by Nelson et al. (1994) including disruption of normal cell function by inhibiting RNA, DNA, and protein synthesis; inhibition of cell division; stimulation of ribotoxic stress response; and activation of mitogen-activated protein kinases. It has been found that T-2 toxin is a potent member of the trichothecene group of mycotoxins produced by *Fusarium* fungi (Bamburg et al.,1970). It has been found that T-2 toxin is a mycotoxin with immunomodulatory activity, where it can stimulate (immune-stimulation) or inhibit (immune-suppression) the activity of the immune system (Shinozuka et al., 1997; Pestka et al., 2004).

To give complete idea about the effect of these collected cases, the internal organs of dead cases during disease outbreak in the same desert districts were subjected for histopathological studies. The results revealed that thickening of the pleural membrane with infiltration of mononuclear inflammatory cells, hemorrhage and proliferation of the epithelial cells lining bronchioles. Moreover, in some cases the proliferation was severe and uncontrolled which lead to occluded the bronchial lumen and form nest of epithelial cells with clear eosinophilic cytoplasm giving the feature of preneoplastic stage (Fig. 1, a & b).Some alveoli were filled with red blood cells accompanied with mononuclear inflammatory cells (alveolar pneumonia). Destruction of the wall of some alveoli with infiltration of inflammatory cells (lymphocytes, macrophages and neutrophils) were noticed accompanied with hemorrhage and calcification (Fig. 2, a & b). Severe hemorrhages with infiltration of inflammatory cells with compensatory emphysema (hemorrhagic pneumonia) were seen in some cases.

While, bronchial lymph node showed moderate to severe depletion of lymphoid follicles, where lymphocytes detected inside alveoli and interalveolar septa. The respiratory tract is the primary route of entry for *Fusarium* spp. and their toxins based on the sinopulmonary involvement. It has been speculated that the fusarium toxins produced damage the tissues which allowing the fungus to spread more easily (Ajello and Hay, 1998). However, Halloy et al. (2005) and (Haschek et al. (2001) mentioned that the lung of experimentally fusariotoxicated piglets particularly with FB1 showed a minimal enlargement of the alveolar septa due to an increase in the macrophage, lymphocyte number and develop lethal pulmonary edema within 4-7 days. Whereas, muscles necrosis and oedema were evident in heart in our study. A various degrees of myocardial degeneration with foci or cellular infiltration and fibrosis were observed in rats with several doses of T-2 toxin, a trichothecene metabolite of *Fusarium* (Schoental et al., 1979).

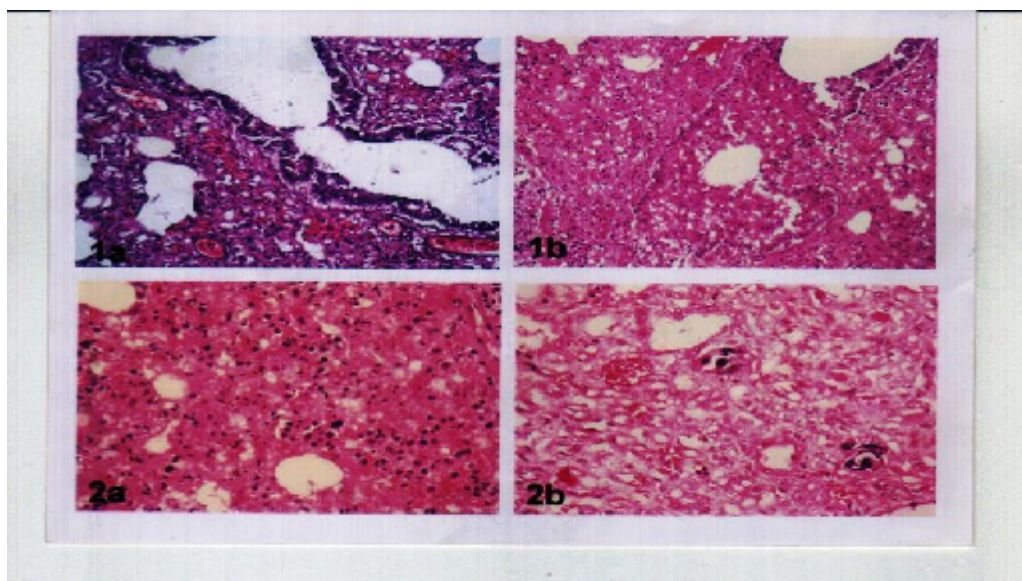


Fig.(1,a & b): Lung of sheep fed on mycotoxin (FB1, T2, ZNE) showing proliferation of the epithelial cells lining bronchiols was severe, uncontrolled and form nest of epithelial cells giving the feature of preneoplastic stage (H & E X 100).

Fig.(2, a & b): Lung of sheep fed on mycotoxin (FB1, T2, ZNE) showing destruction of the wall of some alveoli with infiltration of inflammatory cells (lymphocytes, macrophages and neutrophils) accompanied with hemorrhage and calcification (H & E X a) 200, b) 400).

Many researchers mentioned that fusarium toxins particularly FB1 produces a wide range of biological effects including nephrotoxicity and liver cancer in rats (**Gelderblom et al., 1996**). The present study revealed glissonian's cirrhosis in liver, vacuolar degeneration and necrobiotic changes of hepatocytes in addition to haemorrhages and oedema in between hepatocytes.).Some liver cells arranged in irregular aceni (preneoplastic stage) (Fig. 3 a & b). Epithelial hyperplasia of bile duct was detected with the formation of newly formed bile ductules. There were aggregation of oval vesicular cells in the portal area with infiltration of mononuclear inflammatory cells and fibrous connective tissue formation. Similar lesions were illustrated caused by FB1 (**Abbes et al., 2006 ; Voss et al., 2001**) and zearalenone (**James and Smith, 1982**). According to data of the **National Toxicology Program (USA) (1982)**, ZEN was found to produce hepatocellular adenoma. While, **Abbes et al. (2006)** mentioned that the histological examination of mice kidney that treated with two ZEN doses alone revealed a swelling in the epithelial cells of the proximal tubules, granular degeneration, shrunken glomeruli with the presence of eosinophilic cast in the lumen of tubules and blood vessels dilatation.

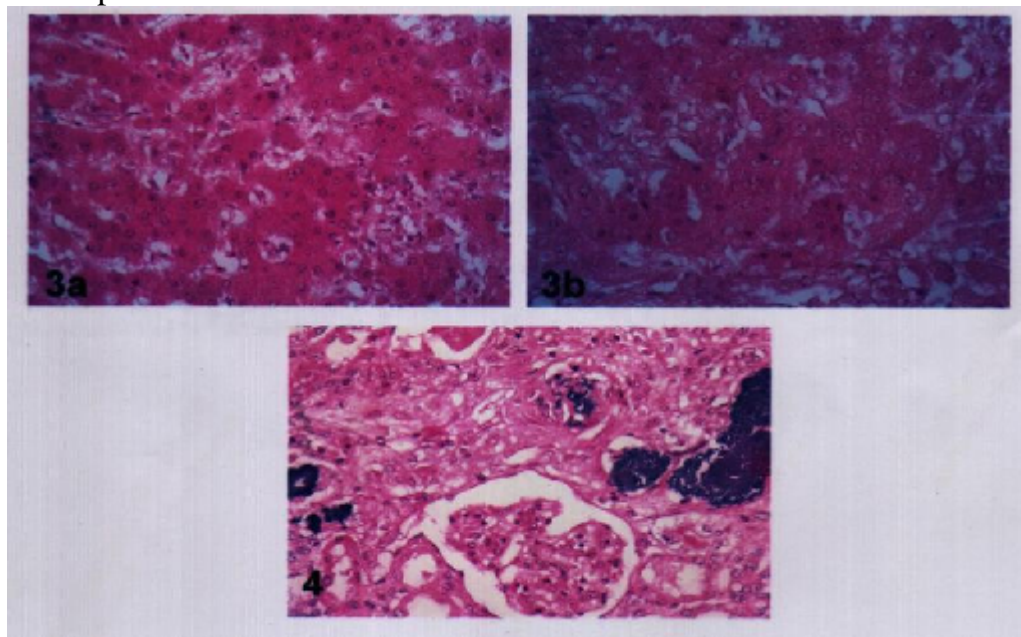


Fig.(3, a & b): Liver of sheep fed on mycotoxin (FB1, T2, ZNE) showing disorganization of hepatic cord (a) with tendency to formation of irregular aceni (preneoplastic stage) (b) (H & E X 400).

Fig. (4): Kideny of sheep feeding on mycotoxin (FB1, T2, ZNE) showing necrosis of renal tubular epithelium, gromerular oedema and calcium deposition. (H & E X 400).

The histopathological examination of kidney revealed dilatation in blood vessels dilatation. Vacuolar degeneration of epithelial cells lining the renal tubules with sloughing in the lumen forming renal casts. Meanwhile, some tubular epithelium revealed necrosis, associated with glomerular oedema and calcium deposition (Fig.4).

Voss et al. (2001), mentioned that FB1 induces apoptosis of hepatocytes and proximal tubular epithelial cells. More advanced lesion in both organs is characterized by simultaneous cell loss (apoptosis and necrosis) and proliferation (mitosis). Microscopic and other findings suggest that an imbalance between cell loss and replacement develops a condition favorable for carcinogenesis. On the molecular level, fumonisins inhibit ceramide synthase and disrupt sphingolipid metabolism and theoretically, sphingolipid-mediated regulatory processes that influence apoptosis and mitosis.

The previous literatures recorded that the pollution affect upon the growth rate and health of human being and animals including aneamia , stunted growth , carcinogenic, tremorgenic, haemorrhagic, dermatitic, pulmonary edema, immunosuppressive and hormonal effects (**Hassan, 1998 and 2003 ; Hassan et al., 2003 ;2004;2008 and 2009**).These findings were confirmed in our study, where , the above results clearly observed the effects of fungal particularly fusarium species and their toxins in sheep at desert districts.

It can induce both toxicologic and immunotoxic effects in a variety of cell systems and animal species as cytotoxic effect to reticulocytes, fibroblasts and lymphocytes and the cellular toxicity appears to be mediated by the inhibition of protein synthesis as reported by (**Ueno, 1983; Rotter et al., 1993; Mogda et al., 2002 ; Hassan et al., 2003 and 2009**). Also, fusarium mycotoxin inhibits cell division, RNA/ DNA synthesis and apoptosis (**Rotter et al., 1996**). Growth retardation and immune suppression are the major toxic effects induced by Fusarium ingestion in farm animals and suppression of the normal immune function

and super induction of pro-inflammatory cytokines have been also suggested as supplementary tools for making a diagnosis as mentioned by (Widestrand et al., 2003 ; Kinser et al., 2004; Hassan et al., 2004). This study, focused the highlight of the dangerous effects of fusarium and their mycotoxins pollution of animal feeds and water which allows a certain generalization as to the solution of problems regarding sheep breeding, which is an important contributor to the country's economy (especially at desert districts) in the form of meat, milk, wool and leather, with respect to the effects of environmental factors.

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